

Supporting Information

A Highly Selective Naphthalimide Based Ratiometric Fluorescence Probe for Recognition of Tyrosinase and Cellular Imaging

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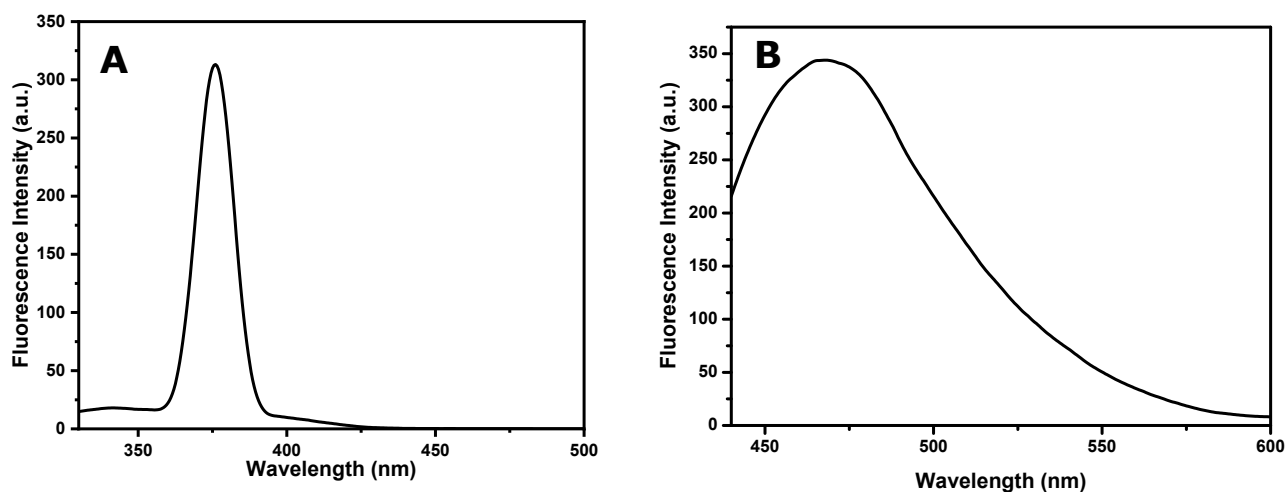


Fig. S1: (A) Fluorescence excitation spectra of probe L3. (B) Fluorescence emission spectra of probe L3 in 10% ACN in PBS ($\lambda_{\text{ex}}=425$ nm, $\lambda_{\text{em}}=467$ nm).

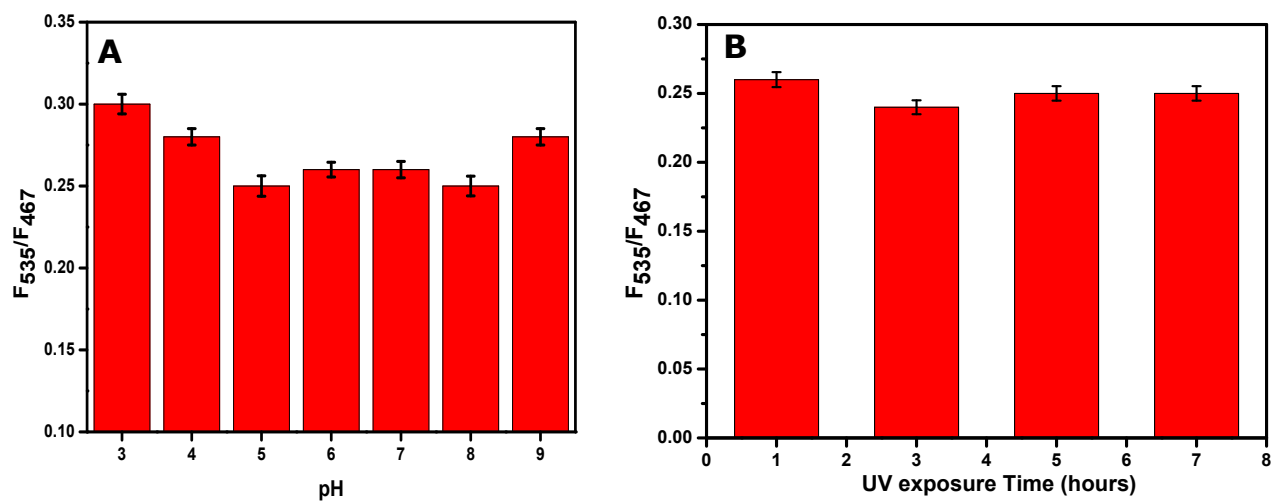


Fig. S2: (A) Fluorescence response (F_{535}/F_{467}) of the probe at different pH. (B) Fluorescence stability study of the probe L3 on exposure to UV light at a different time interval.

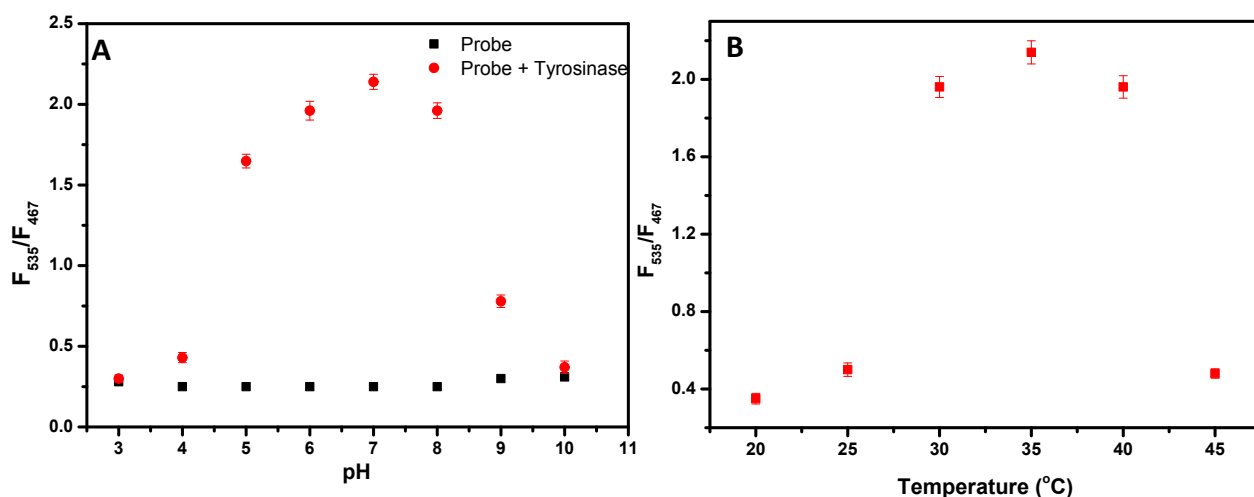


Fig. S3: (A) Fluorescence response of probe in the presence of tyrosinase (150 U mL⁻¹) at different pH. (B) Effect of temperature on fluorescence response of probe in the presence of tyrosinase (150 U mL⁻¹).

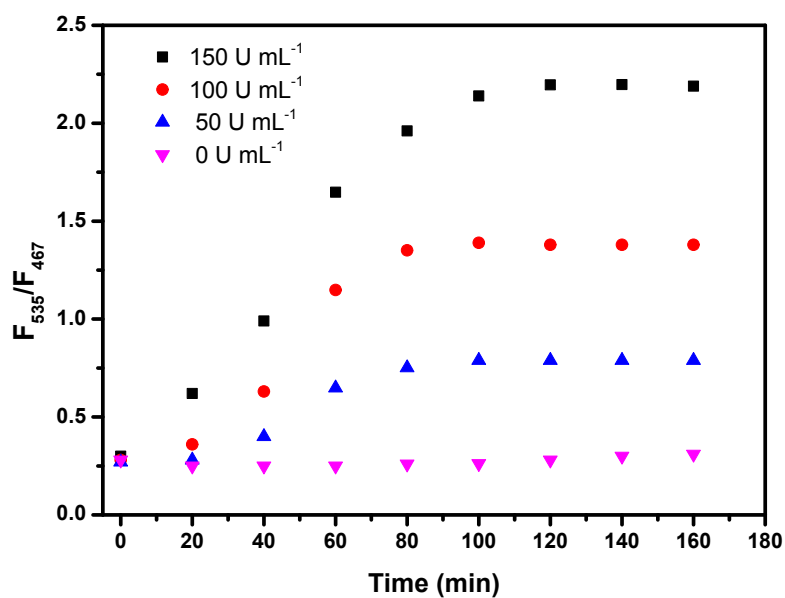


Fig. S4: Fluorescence emission plot of probe L3 (10 μM) vs. reaction time (0-160 min.) at different concentration of tyrosinase (0- 150 U mL⁻¹). Experiments were performed at 37 °C in PBS (10% ACN pH 7.4) with $\lambda_{ex} = 425$ nm.

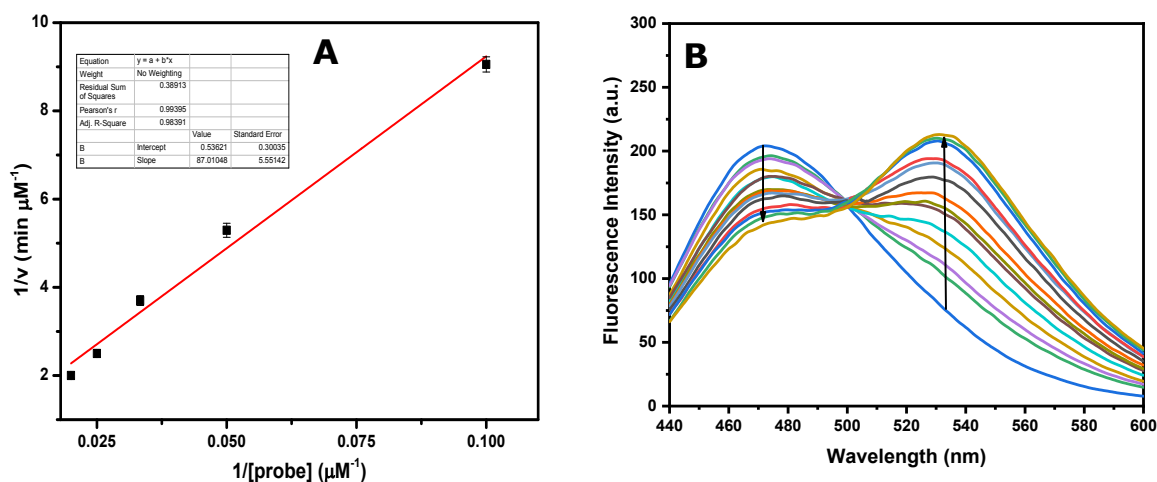


Fig. S5: (A) Kinetic parameter study for the calculation of K_m . (B) Fluorescence emission spectra of Probe L3 in response to tyrosinase in 1:99 (v/v) DMSO/PBS.

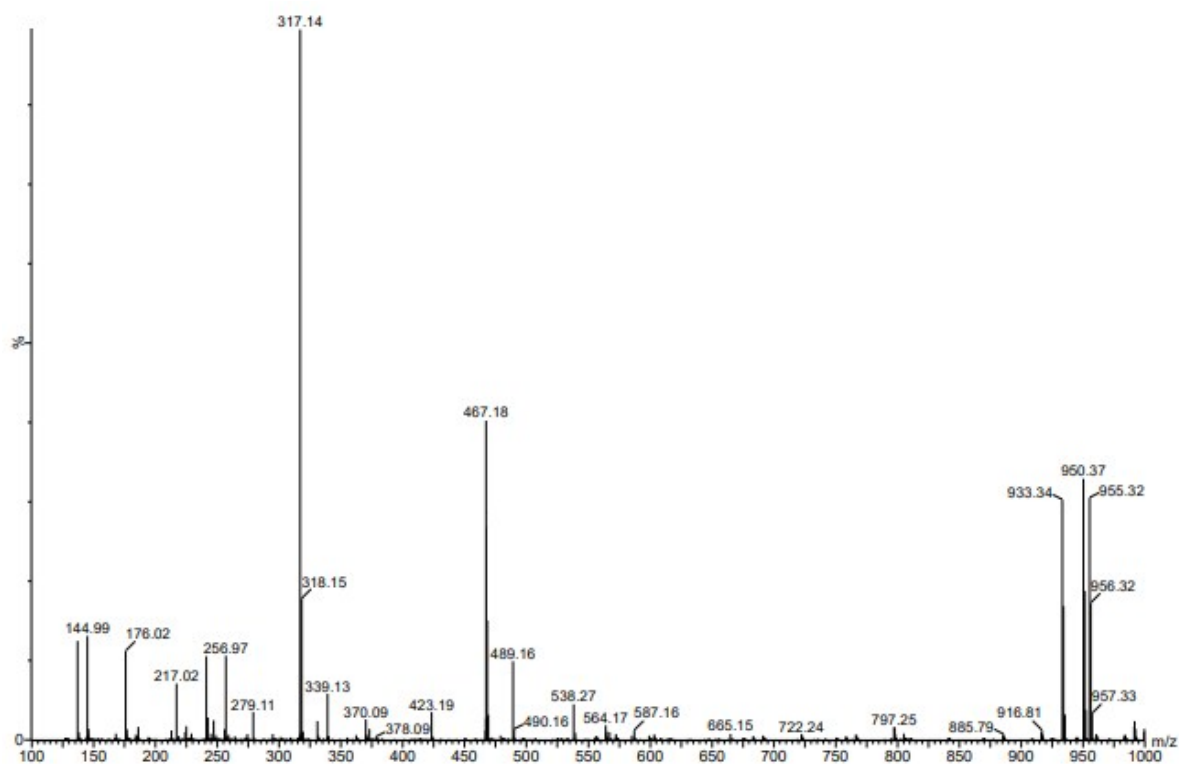


Fig. S6: Mass spectra of L3 solution in the presence of tyrosinase. Peak correspondence to m/z at 317.14 indicate the breakage of carbamate linkage and release of free 4-aminonaphthalimide derivative (L2).

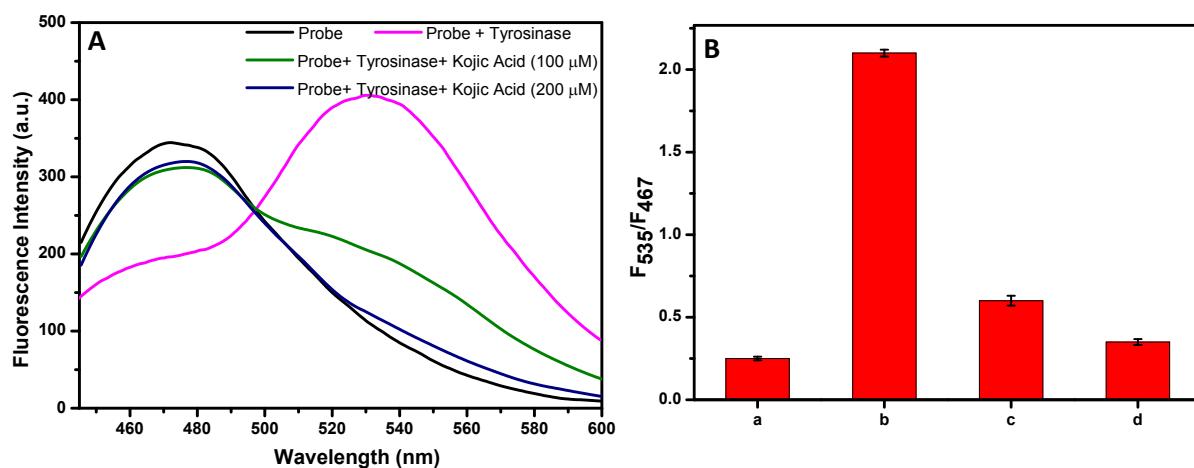


Fig. S7: (A) Fluorescence emission profile of probe at different reaction conditions. (B) Fluorescence intensity ratio of probe at F_{535}/F_{467} . (a) Probe (b) Probe + Tyrosinase (c) Probe + Tyrosinase + Kojic acid (100 μM) (d) Probe + Tyrosinase + Kojic acid (200 μM)

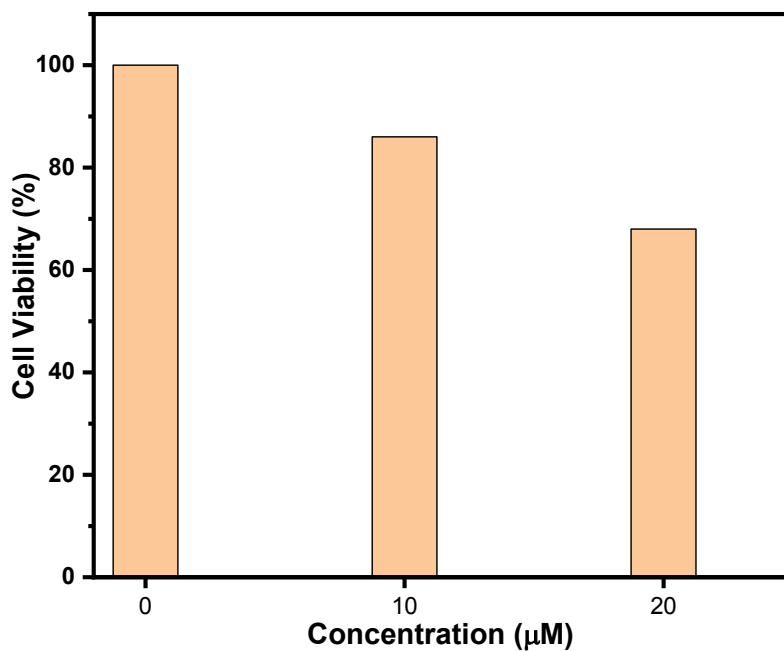


Fig. S8: Cytotoxicity Assay of Probe L3.

Table. S1: Fluorescence sensor for the detection of Tyrosinase with a different detection limit

Method	Limit of Detection	Reference
Nanoclusters of gold	6 U L ⁻¹	1
RF-QDs-DA	10 U L ⁻¹	2
Cyanine	0.01 U mL ⁻¹	3
Pdots@Tyr-OMe	1.1 U L ⁻¹	2
Dopa-CQDs	17 U L ⁻¹	2
CDs-Tyr	10.2 U mL ⁻¹	4
Resofuran	0.04 U mL ⁻¹	5
Naph-L3	0.2 U mL ⁻¹	This work

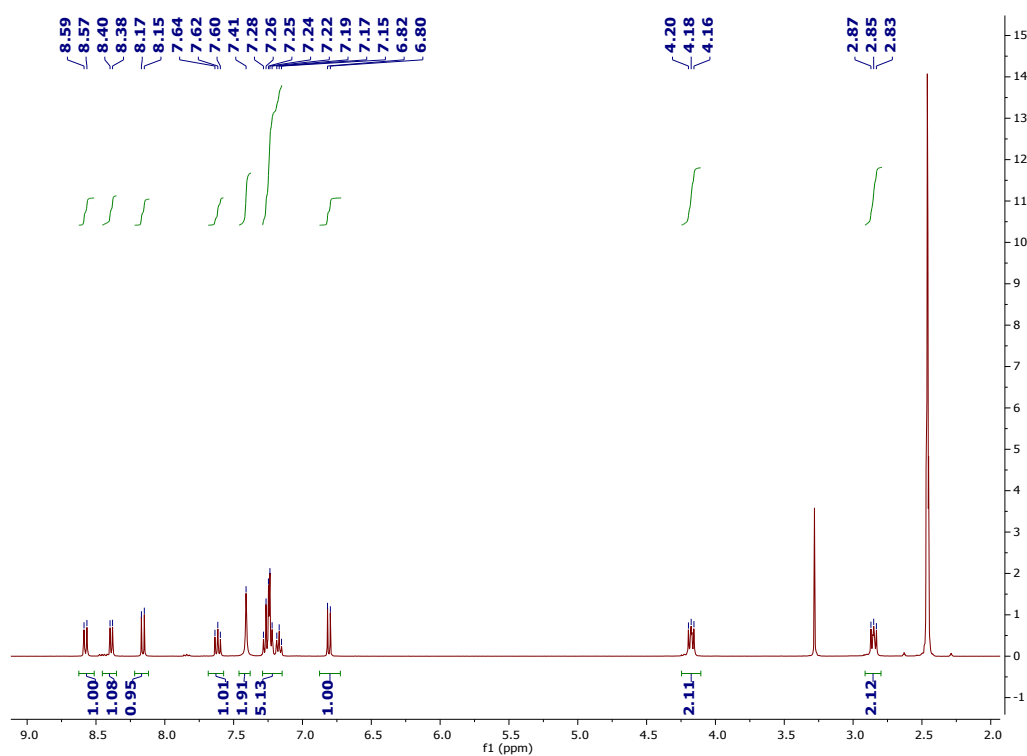


Fig. S9: ¹H NMR of L2

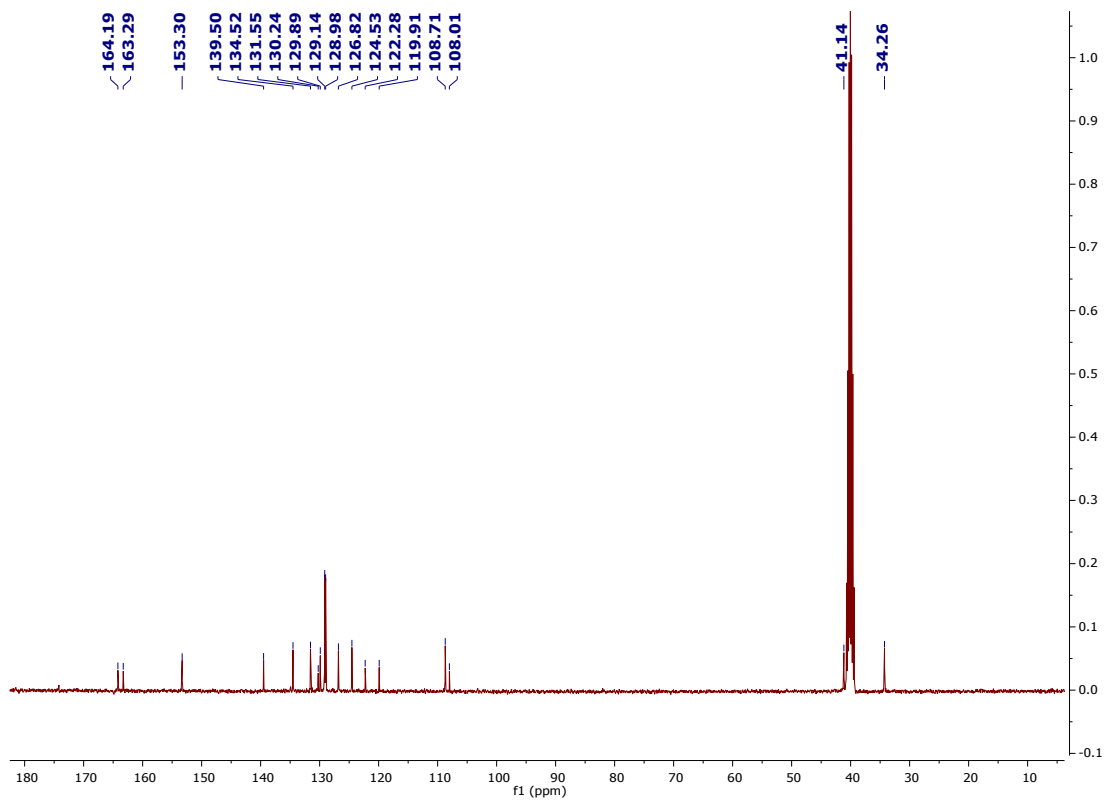


Fig. S10: ^{13}C NMR of L2

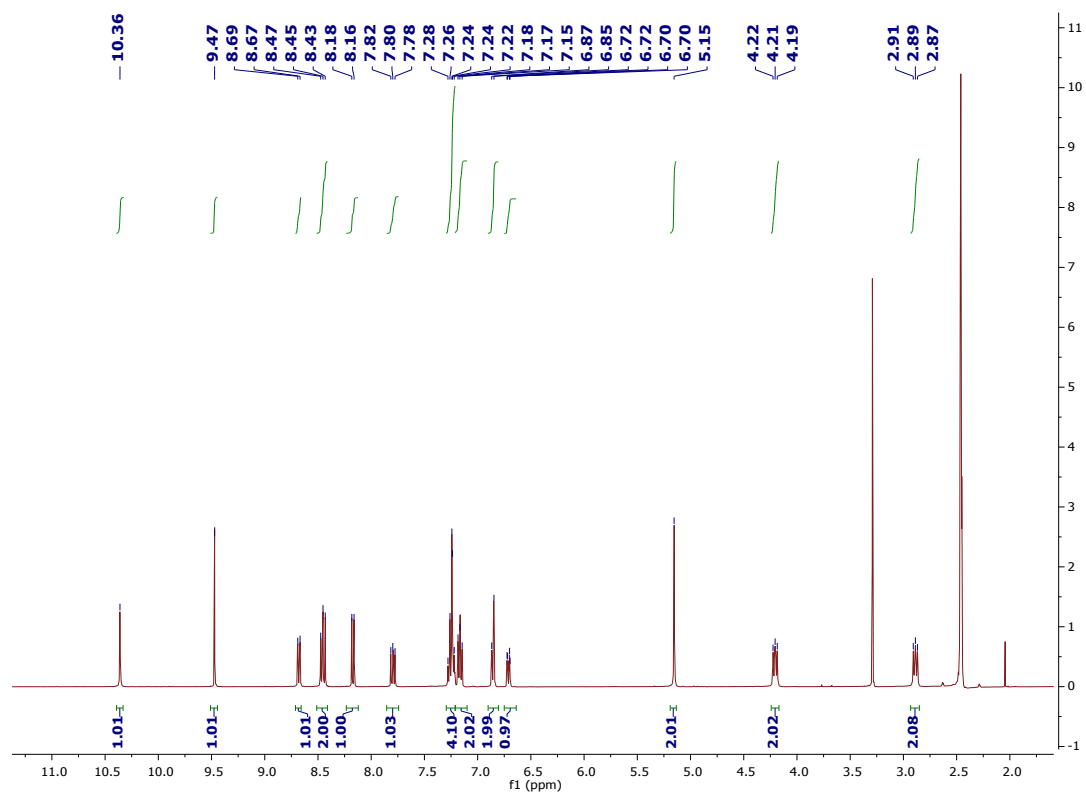


Fig. S11: ^1H NMR of Probe L3

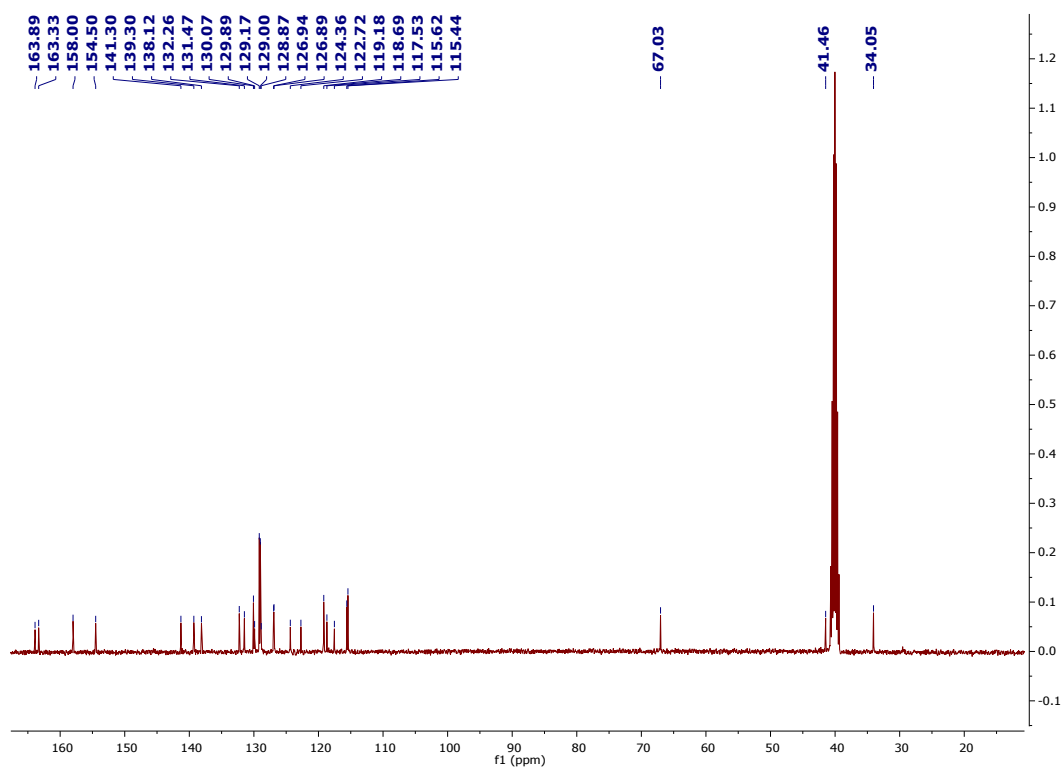


Fig. S12: ^{13}C NMR of Probe L 3

Single Mass Analysis

Tolerance = 15.0 PPM / DBE: min = -1.5, max = 50.0

Element prediction: Off

Number of isotope peaks used for i-FIT = 3

Monoisotopic Mass, Even Electron Ions

5 formula(e) evaluated with 1 results within limits (up to 50 closest results for each mass)

Elements Used:

C: 25-30 H: 15-24 N: 0-2 O: 0-5

Sample Name : S-271

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XEVO G2-XS QTOF

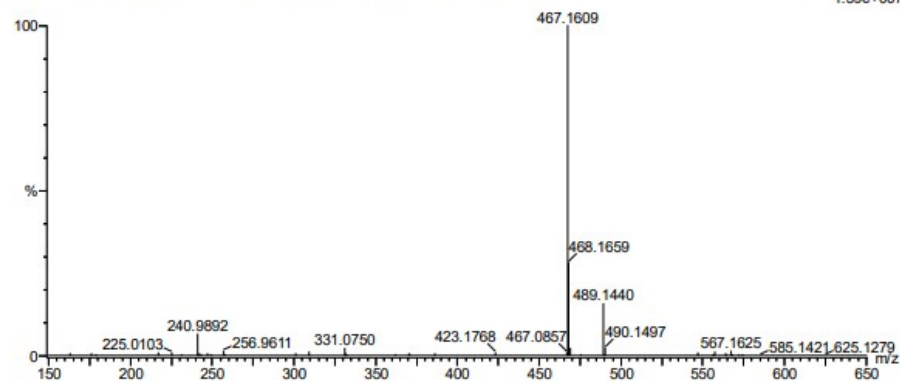
Test Name : HRMS-1

ROPAR

1: TOF MS ES+

060418-S-271 9 (0.105) AM (Top,4, Ar,10000.0,0.00,0.00); Sm (Mn, 1x3.00); Cm (7:16)

1.59e+007



Minimum:

Maximum: 5.0 15.0 -1.5

Mass	Calc. Mass	mDa	PPM	DBE	i-FIT	Norm	Conf(%)	Formula
467.1609	467.1607	0.2	0.4	18.5	495.7	n/a	n/a	C28 H23 N2 O5

Fig. S13: HRMS of Probe L3

References:

1. Y. Teng, X. Jia, J. Li and E. Wang, *Anal. Chem.*, 2015, **87**, 4897-4902.
2. J. Sun, H. Mei, S. Wang and F. Gao, *Anal. Chem.*, 2016, **88**, 7372-7377.
3. X. Li, W. Shi, S. Chen, J. Jia, H. Ma and O. S. Wolfbeis, *Chem. Comm.*, 2010, **46**, 2560-2562.
4. J. S. Sidhu and N. Singh, *J. Mat. Chem. B*, 2018, **6**, 4139-4145
5. X. Wu, X. Li, H. Li, W. Shi and H. Ma, *Chem. Comm.*, 2017, **53**, 2443-2446.